

## WEST Search History

DATE: Friday, November 28, 2003

<u>Set Name</u> side by side	<u>Query</u>	<u>Hit Count</u>	<u>Set Name</u> result set
<i>DB=USPT,PGPB,JPAB,EPAB,DWPI; PLUR=YES; OP=ADJ</i>			
L12	L10 and (transgenic (mouse or mice))	11	L12
L11	L10 and (transgenic animal)	17	L11
L10	L9 and (transcription complex)	17	L10
L9	L8 and complex	69	L9
L8	L7 and transgenic	73	L8
L7	L5 or L6	107	L7
L6	L2 and (epitope near tagged)	48	L6
L5	L2 and (epitope near tag)	76	L5
L4	L2 and (epitope near tag\$)	50	L4
L3	TBP	2499	L3
L2	TBP and TATA	371	L2
L1	TATA-box binding protein	28	L1

END OF SEARCH HISTORY

Amz  
11/28/03

?ds

Set	Items	Description
S1	3995	TATA(W) BOX(W) BINDING(W) PROTEIN
S2	32	S1 AND TRANSGENIC
S3	21	RD (unique items)
S4	7417	TBP AND TATA
S5	86	S4 AND TRANSGENIC
S6	3	S5 AND EPITOPE

?t 3/3,ab/1-21

Dialog  
file:medicine  
11/28/03  
Amz

6/3,AB/1 (Item 1 from file: 34)  
DIALOG(R) File 34:SciSearch(R) Cited Ref Sci  
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04682540 Genuine Article#: UB729 Number of References: 84

**Title: DROSOPHILA TFIID BINDS TO A CONSERVED DOWNSTREAM BASAL PROMOTER  
ELEMENT THAT IS PRESENT IN MANY TATA -BOX-DEFICIENT PROMOTERS** (  
Abstract Available)

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Corporate Source: UNIV CALIF SAN DIEGO, DEPT BIOL/LA JOLLA//CA/92093; UNIV  
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**Abstract:** We describe the identification and characterization of a conserved downstream basal promoter element that is present in a subset of *Drosophila* **TATA** -box-deficient ( **TATA** -less) promoters by using purified, **epitope** -tagged TFIID complex (eTFIID) from embryos of **transgenic** *Drosophila*. DNase I footprinting of the binding of eTFIID to **TATA** -less promoters revealed that the factor protected a region that extended from the initiation site sequence (about +1) to similar to 35 nucleotides downstream of the RNA start site. In contrast, there was no apparent upstream DNase I protection or hypersensitivity induced by eTFIID in the -25 to -30 region at which **TATA** motifs are typically located. Further studies revealed a conserved sequence motif, (A)/(G)G(A)/(T)CGTG, termed the downstream promoter element (DPE), which is located similar to 30 nucleotides downstream of the RNA start site of many **TATA** -less promoters. DNase I footprinting and in vitro transcription experiments revealed that a DPE in its normal downstream location is necessary for transcription of DPE-containing **TATA** -less promoters and can compensate for the disruption of an upstream **TATA** box of a **TATA** -containing promoter. Moreover, a systematic mutational analysis of DNA sequences that encompass the DPE confirmed the importance of the consensus DPE sequence motif for basal transcription and further supports the postulate that the DPE is a distinct, downstream basal promoter element. These results suggest that the DPE acts in conjunction with the initiation site sequence to provide a binding site for TFIID in the absence of a **TATA** box to mediate transcription of **TATA** -less promoters.

6/3,AB/2 (Item 1 from file: 266)

DIALOG(R) File 266:FEDRIP

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IDENTIFYING NO.: 0185997 AGENCY CODE: AGRIC

**Regulation of Trans-Gene Expression in WheatSDTDBT**  
plant development

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**SUMMARY:** Our objectives are as follows. Using our wheat transformation system we plan to (1) determine how expression levels of an introduced transgene, wheat **TATA** -Binding Protein ( **TBP** ), affect wheat suspension cells and plants; and (2) use **epitope** -tagged wheat **TBP** to identify and isolate the multi-polypeptide complexes that contain it, such as those including **TBP** Associated Factors (TAFs) and holoenzyme. To improve plant characteristics by genetic engineering, we must understand regulatory mechanisms of gene expression. Our previous studies used reconstituted in vitro systems to examine transcriptional regulation. We will now analyze regulation in vivo so we can rationally approach

transgene design to ensure correct spatial and temporal expression, prior to modifying plant traits. This approach requires that we have a method to deliver trans-gene DNA to prepare **transgenic** wheat. A second

purpose for the in vivo system is the introduction of **epitope** -tagged proteins into wheat in order to isolate regulatory complexes. The **epitope** tag allows for efficient biochemical isolation of these complexes, which other wise must be purified by conventional biochemical means, resulting in poor yields and possible inactivation. One of the most efficient delivery gene methods for dicots is binary vector-Agrobacterium-mediated gene transfer, which favors high

frequency integratiion into the host genome, in low copy number and in appropriate chromatin isochores. When it was observed that actively dividing tissue and the phenolic acetosyringone permitted Agrobacterium-mediated transformation in the monocot rice, we developed a similar method for wheat (*Triticum aestivum* cv chinese Spring). We used immature wheat embryos and embryogenic callus, because they allow for high transformation rates, and Chinese Spring has a very high regeneration frequency. We achieved a stable transformation rate of 1-2%, better than earlier studies and comparable to biolistics. Our first objective is to determine how the transferred wheat **TBP** gene

affects growth and development of **transgenic** callus, and then **transgenic** wheat suspension cells. Regeneration of plants will permit the further analysis in a whole organism. The second objective will be to initiate studies to better isolate native polypeptide complexes containing **TBP**, and to validate in vitro gene regulation models in an in vivo system. To determine how **TBP** expression levels affect plant cells. Our transformed wheat callus expresses **epitope** (FLAG-(His)6 domain)- **TBP**. We will next determine if **epitope** - **TBP** is expressed in excess of endogenous wheat **TBP**. **Epitope** - **TBP** will be removed from cell extracts and quantitated by anti-FLAG antibody and Talon affinity

chromatography resins, and then endogenous **TBP** quantitated by our anti- **TBP** antibody affinity resin. S1 nuclease RNase protection, primer extension, and/or PCR analysis will quantify transgene mRNA levels, using a DNA probe to the FLAG-(His)6 domain to discriminate **transgenic** from endogenous **TBP**. These data will indicate the levels of **epitope** - **TBP** expression. We will assay transient expression first in transformed callus and then in transformed wheat suspension cells. Geneticin selection of transformed suspension cells will produce homogeneous cell lines of stably transformed cells. We will also look for phenotypic effects on transformed plant cells compared to those transformed with a

control vector without wheat **TBP**, or our truncated, non-functional form of **TBP**. Phenotypic measures will include cell viability (dye exclusion) and growth rate (cells/ml). As **transgenic** plants become available we will continue the gene expression studies and non-invasively measure developmental effects as leaf lengths, plastochron lengths (time interval to make each leaf), floral transition time etc. We may observe that with more **TBP** there is more transcription. A second possibility is that gene regulation will be so deranged as to be lethal. Phenotypic studies and mRNA levels during

6/3,AB/3 (Item 1 from file: 399)

DIALOG(R) File 399:CA SEARCH(R)

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130048272 CA: 130(5)48272v PATENT

Purification of higher order transcription complexes from epitope-tagged TATA box-binding protein-expressing transgenic non-human animals

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LOCATION: Germany,

ASSIGNEE: Hoechst Aktiengesellschaft

PATENT: European Pat. Appl. ; EP 881288 A1 DATE: 19981202

APPLICATION: EP 98109516 (19980526) \*EP 97108433 (19970526)

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DESIGNATED COUNTRIES: AT; BE; CH; DE; DK; ES; FR; GB; GR; IT; LI; LU; NL; SE; MC; PT; IE; SI; LT; LV; FI; RO

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